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I.—On a Uredine affecting the Himalayan Spruce-fir (Abies smithiana, Forbes).—By SURGEON A. BARCLAY, M. B., Bengal Medical Service.

[Received December 20th ;--Read January 6th, 1886.]

(With Plates I., II., and III.)

During a short holiday spent in the interior of the Western Himalayas beyond Simla during May, I observed many fir trees (*Abies smithiana*) attacked by a conspicuous parasite which, upon examination, proved to be a uredine—and apparently a new one, for its characters do not correspond with those of any other that I am acquainted with occurring on the *Coniferae*. It occurred in great abundance throughout the forests of the Sutlej valley at elevations of from 7,000 to 10,000 feet. I have not been able to obtain any evidence of its extension eastwards along the Himalayas,* but in a westerly direction Colonel Collett informs me that he has met with it in the Kuram Valley of Afghánistán. He unfortunately did not preserve any specimens, but, from his description of the affection as he saw it there, I think there can be little doubt about its identity.

I have no data to enable me accurately to limit the season during which the parasite continues to be outwardly recognisable. It was met with in various stages of development throughout May, and I believe it

* See note at end of paper.

[No. 1,

disappears entirely during the rains in July. Whether the two forms in which it was met with, and which I shall for convenience describe as the æcidial and uredinal, do really stand to one another in this relationship it is impossible to determine without experimental evidence, and I have had no opportunities for performing the necessary experiments. From other circumstances, however, the existence of such relationship appears to be probable. I could find no trace of any teleutosporic form for this parasite either on the same or on any other host. Although I examined many uredo pustules with care, I could never detect any separate form of persistent spore among them. This, however, may have been due to the fact that all the specimens at my disposal for investigation were gathered during May, and, if teleutospores are formed at all in the uredo beds, it is quite possible and probable that they are developed later. The question as to whether the mycelium in either form is perennial requires further investigation. I am mainly influenced in regarding the mycelium in both instances as not perennial by the fact that the affection in both cases is confined entirely to shoots of the present season's growth, as will be more fully explained below, and that the whole of the affected shoot withers and dries up completely when the fungus has completed its existence (fig. 20).

In describing these two forms of the affection, it will be convenient in the first place to give a short account of the general naked eye appearances of each, and then to proceed with the description of their minute characters.

General Appearance of the Æcidial Affection.—When fully developed the affection is very conspicuous : large masses of tissue are affected, and the pale yellow colour of these parts contrasts strikingly with the surrounding green foliage. The fungus always attacks a young terminal shoot, and the distortion caused, partly a real and partly a pseudo-hypertrophy, renders it still more conspicuous. Moreover; the affected part assumes a drooping habit due apparently to the weakening of the texture of the stem by the mycelium which largely pervades its whole tissue.

The general appearance of a young affected shoot is well shown in fig. 17. In this stage, the affected needles lie against one another, embracing the stem instead of standing out from it, as the natural needles are seen to do in the same figure. The stem and needles are already in this young stage considerably thickened. The whole of the affected part has a uniformly pale yellow colour. Conspicuous as it already is, it becomes much more so when further advanced. The thickening of the needles and stem increases greatly. The dimensions of the transverse section of a normal needle on an average are 1886.7

The length of a fully affected shoot varies considerably, from an inch or two to 12 inches or more. In one specimen, a terminal affected shoot measured 9 inches in length, whilst two lateral shoots, also affected and springing from its base, measured $6\frac{1}{2}$ and 5 inches respectively (fig. 1). The measurements of another very fine specimen, gathered from a branch near the summit of a young tree about 15 feet high, were as follows. The small branch upon which the affected shoots occurred sprang immediately from a main lateral branch. This branch contained 15 shoots in all. Their lengths beginning from the base were as follows :-- the first 1 inch, the second $1\frac{1}{2}$ inches, the third 2 inches, the fourth and fifth 3 inches, and the sixth $3\frac{1}{2}$ inches. So far all the shoots were quite free of the affection, but the seventh was affected and measured 5 inches, the eighth unaffected $4\frac{1}{2}$ inches, the ninth affected 9 inches, the tenth unaffected $5\frac{1}{2}$ inches, the eleventh affected 8 inches, the twelfth unaffected $6\frac{1}{2}$ inches, and the fifteenth and last affected 12 inches. Thus the affected shoots were on the whole considerably longer than the unaffected. I cannot explain the curious phenomenon here displayed of alternate shoots only becoming affected and with such regularity. The phenomenon becomes even more striking when it is observed that not a single needle on an affected shoot escapes invasion, whilst, on the contrary, no single needle on one of the intervening unaffected shoots, nor any one on the main stem, even in the internodes between affected shoots, is ever attacked in any degree. To all outward appearance, the disease is confined absolutely to shoots of the present season's growth and does not extend to the branch of which it is an immediate shoot, and which was of the preceding season's growth.

As regards the general characters and appearance of the affected needles, they are, as already stated, considerably thickened and curved (fig. 2). The distal end of the needle is densely studded with spermagonia, which appear as minute dark points uniformly distributed all around it. Towards the middle half these become much less numerous, and, as the base is approached, they are few and confined to the inferior surface of the needle. The spermagonia are developed considerably earlier than the æcidia. The general appearance of the æcidia is illustrated in figs. 2 & 3. They are borne mainly on the upper surface of the spine in two parallel rows. In rare cases, they burst through the inferior surface, but such æcidia are isolated and smaller than those on the upper surface (fig. 2 a). While still unopened, the

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secidium is of a pale rose colour, but the free spores in mass are orangered, whilst the peridium is colourless. The accidia are first developed on the lowest needles of an affected shoot, whilst in the upper ones they appear in succession from below upwards. They are generally more numerous on the lower than on the upper needles of a shoot. The still unopened æcidia protrude through clean rents in the epidermis, and, when ripe, the peridium bursts with an irregular frayed margin (figs. 3 & 4).

General Appearance of Uredinal Affection.-The uredinal form is much more frequently met with than the æcidial just described. Its general appearance is shown in fig. 19, which represents a fully developed specimen. This specimen had been preserved for a few days in brine, and the needles were swollen somewhat unnaturally. The dimensions of a fully affected needle are 1.60×1.43 m.m., thus exceeding those of the æcidial affections. This form occurs sometimes on the same host which bears the æcidial form, but more frequently a separate tree. Presuming that the uredinal form is genetically related to the æcidial, this would not be surprising, since æcidial fructification is usually less frequently developed than the uredinal. Whenever the æcidial form is met with, it is rarely unaccompanied by the uredinal on the same host. In their general appearances alone, the two affections, when fully developed, differ so much as to enable a distinction to be made between them at a glance (compare figs. 18 & 19). The densely aggregated needles of the uredinal form contrast strikingly with the opener habit in the æcidial form. But, in addition to this difference, the uredinal form is distinguishable by its orange-red colour. It resembles the æcidial affection in always being confined to shoots of the present season's growth, but the affected shoots are always small and never attain the lengths so often seen in the æcidial form. Indeed, the shoots affected by the uredinal form of the parasite appear to be stunted in growth, since no affected shoot ever measures more than 2 inches in length, whilst in the measurements of unaffected shoots (given above in contrasting them with neighbouring shoots affected by the æcidial form) many exceeded this length. The aggregation of the needles also points to a suppression in the long growth of the stem. It is probable, therefore, that, whilst the acidial parasite stimulates growth in length of the stem, the uredinal one represses it. A specimen gathered from the same host which bore the æcidial affection, and whose measurements were first given above, measured only 2 inches in length though fully developed. The separate needles here also curl up considerably, contrasting strikingly with the neighbouring erect unaffected needles (fig. 19). Every needle of the affected shoot is involved.

4

1886.] Himalayan Spruce-fir (Abies smithiana).

When mature, the needles are clothed densely with a homogeneous orange-red layer of spores forming elevated beds. These masses of spores break out first in isolated spots along the lateral grooves on the upper surface of the needle. Having once broken through the epidermis, they extend in area with great rapidity and soon coalesce. Ultimately, they form two longitudinal beds on the upper surface separated by a narrow groove in the middle line. Laterally, they creep round and involve the lower surface to some extent. After the spores have burst through the upper surface and have made some progress towards coalescence, isolated crops appear in two more or less parallel lines on the lower surface of the needle. These ultimately form oval beds which do not tend to coalesce as on the upper surface. When quite young, this uredinal form emits a disagreeable and offensive odour which is absent from the æcidial form.

Microscopic Characters. (a) Normal Structure of Needle.-The normal needle is flattened laterally, and a transverse section is broadly quadrilateral (fig. 21). The extreme length of such a section about the middle of the needle is about 1.09 m.m. and the breadth 0.76 m.m. The two sides forming the upper angle are shorter than the two lower sides, and include a larger angle. On either side, near the surface and about midway between the upper and lower angles, are two resin canals, and in the centre is a vascular bundle measuring 0.236 m.m. in diameter. The needle is covered externally by a layer of epidermis cells whose continuity is interrupted only by the interposition of stomata. In each transverse section, generally six stomata may be seen, two on each of the shorter sides and one on each of the longer ones. Immediately under the epidermis, there is a layer of thick-walled hypodermal cells. This consists of a single row of cells, excepting at the lower angle, where it is double. It is interrupted at the stomata and at the resin canals. These canals are bounded by two layers of cells; the external larger and thicker-walled than the internal. The canals measure about 0.076 m. m. in diameter. The centre of the needle is occupied by the vascular bundle, which in transverse sections is seen to be limited by a circle of oval cells forming an endodermic sheath. Between these sheath cells and the hypoderma, lie large thin-walled parenchyma cells (the mesophyll) with numerous large air spaces. These parenchyma cells, as seen in transverse section, resemble, to some extent, the palisade cells of ordinary leaves and are arranged radially; but this general radial symmetry is interrupted by a secondary crescentic system around each of the resin canals. In depth, these cells vary from two to four cells. They are filled with chlorophyll corpuscles, starch grains, and protein bodies, the relative amounts varying with the season and

age of the needles. The sheath cells are considerably longer than they are broad. They do not contain chlorophyll but often large starch grains. Within the endodermic sheath are first thin-walled parenchyma cells, with sinuous outlines opposite the xylem, but more cubical opposite the phlœm. These cells have large bordered pits. The xylem is towards the upper side and the phlœm below it. The xylem is more or less distinctly divided into two lateral halves by two to three or more large thick-walled sclerenchymatous cells. The phlœm is more distinctly divided into two lateral halves. Beyond the phlœm is a group of from three to seven large thick-walled sclerenchyma cells, and, in the centre of this, immediately adjoining the phlœm, one large empty thin-walled cell may always be seen.

(b) Microscopic Characters of the Æcidial Fungus.-The general appearance of the invaded tissue of a needle in a transverse section is well shown in fig. 22, and, if this be compared with fig. 21 (exhibiting a transverse section of a normal needle), the general deviations in the structure of the former from the normal condition are at once apparent. The parenchyma cells are larger and rounder than in the normal condition. They measure from 0.177 or 0.101 m.m. in length by 0.058 in width, whilst in the normal needle these dimensions are from 0.093 or 0.071 by 0.035 m.m. The radial arrangement of these cells is also entirely lost, and no vestige of the resin canals remains. The most striking change, however, produced in the invaded tissue is the separation or dislocation of the parenchyma cells from one another by the mycelium. In every transverse or longitudinal section of a needle, it is easy to trace the course of the mycelial filaments for a short distance in the parenchymatous tissue (figs. 5 & 6). In these figures, it will be observed that, though both are magnified equally, the hyphæ of the one are greater in diameter than those of the other. The cause of this difference is due to their different treatment and mounting. Fig. 6 represents a section which was stained in picrocarmine and immediately mounted in Farrant's gum solution, whilst the section represented in fig. 5 was stained with vesuvin, then immersed in absolute alcohol, cleared in clove oil, and mounted in canada balsam. I had no opportunity of examining the tissues when they were quite fresh, and the following measurements of the diameter of the hyphæ are therefore subject to correction. The diameter of the filaments illustrated in fig. 6 (mounted in gum) was on an average 4.7μ , whilst the average diameter of those represented in fig. 5 was $3 \cdot 1\mu$. The mean of these two measurements would probably very nearly approach a correct measurement of the fresh filaments, for, whilst the former measurement may be a little excessive from absorption of water, the latter is probably reduced by immersion in alcohol. In many places, the mycelial filaments may be seen in parallel lines closely applied to the external surfaces of the parenchyma cells. The main mass of the mycelium, therefore, lies in the parenchymatous tissue, that is, between the hypodermal cells and the endodermic sheath surrounding the central wood bundle. A few filaments may be seen insinuated between the hypodermal cells and reaching the epidermis cells. The extension of the mycelium inwards is largely and conspicuously arrested at the endodermic sheath. Some filaments do, however, pass between the cells of this sheath and may be traced among the parenchyma cells. A very few may be traced in the phlœm, but none in the xylem. A special aggregation of filaments occurs between the phlœm and the cells of the sheath, entirely replacing the central sclerenchyma cells.

The minute structure of the mycelial filaments does not present any special characteristics (fig. 9): they are septate and branched. While coursing through the parenchyma, they are not specially convoluted, but become very much so at the bases of the æcidia and spermagonia. A few haustoria occur in the cells of the parenchyma: they are of the branched type and by no means numerous: as the mycelial filaments are so closely applied to the cell walls, appearing as if cemented to them, there would appear to be little necessity for haustoria.

My specimens were unfortunately not favourable for the investigation of the course of the mycelium in the stem. The mycelial filaments were, however, seen to ramify abundantly both in the cortical tissue and also among the pith cells. The circle of vascular tissue was frequently found to be broken in outline in order to allow of strands of the filaments entering. I was unable to detect any actually between the wood cells. The filaments here presented the same characters as those described above.

Spermagonia.—These, as already stated, occur in great numbers; they are deeply set with their bases beneath the hypoderma, and measure about 0.139 m. m. in length and breadth. The conical neck protrudes 45μ above the level of epidermis (fig. 12). They are of the usual structure and require no description. The mycelium at their bases forms densely convoluted masses which thrust the epidermis and hypodermal cells widely apart. The spermatia are very minute oval bodies (fig. 10).

Æcidiospores.—By reflected light the æcidiospores are pale yellow. They are long irregularly oval bodies densely beset externally with minute spines or tubercles. Many of them are rounder at one extremity than at the other and some are flattened and curved (fig. 7). The dimensions of the dry spores were on an average 38μ by 16μ . These measurements were on the whole very uniformly maintained. After the spores had been moistened with water for a few minutes, their average length became 44μ and their average breadth 21μ . The epispore is thick, measuring about $2\cdot 1\mu$. Seen in optic section, the epispore appears to be coarsely radially striated, an effect due probably to the overhanging and underlying processes on the surface. The endospore appears to consist of a fine homogeneous membrane, but is ill-defined. The contents are finely granular and yellow (fig. 7).

The spores are given off serially in rows from the hymenium, a small intercalary cell separating successive spores (fig. 8.) Traced from without inwards each successive spore becomes smaller, until at last no distinction can be made between spore and intercalary cell, and these merge gradually into the round basidial cells forming the floor of the hymenium.

The peridium is very resistant and consists of two layers of cells which are considerably larger than the spores (fig. 16).

(c) Microscopic Characters of the Uredinal Fungus.—The general distribution of the mycelium in the tissue of the needle is shown in figs. 23 and 25, which represent transverse sections. Masses of mycelial filaments occur between the cells of the parenchymatous tissue. The dislocation of the cells by the mycelium is much more uniform and symmetrical than in the case of the æcidial affection, where special aggregations are apt to occur at certain places. The filaments in this form are also much more convoluted. Both in transverse and longitudinal sections the masses of mycelium present the appearance of confused more or less circular outlines resembling a pseudoparenchyma. This appearance is due to the extreme convolution of the filaments and to their frequent branching (fig. 14). The majority of the hyphæ have a greater diameter than those of the æcidial form; but they vary in this respect greatly. Most of the hyphæ measure about 8μ in breadth, but some are to be seen of half that size (fig. 13, a § b.)

In this form, as in the last, the great mass of the mycelium is confined to the parenchymatous tissue. The arrest of the filaments at the sheath enclosing the central wood bundle is very abrupt, more so even than in the æcidial form, for the continuity of the cells is scarcely at all broken (fig. 25). Some filaments, however, do pass through and may be traced both between the parenchymatous cells and to a lesser extent within the phlœm. In this form, also, the central sclerenchyma cells are replaced by a mass of convoluted mycelium. In every transverse section, the single large empty cell noted in the description of the normal needle may be seen between this mass of mycelium and the phlœm. Elsewhere within the central bundle, the hyphæ are less numerous and arranged 1886.]

in a more or less scalariform manner. In this uredinal form, the mycelial filaments do not closely encase the parenchyma cells, as they were shown to do in the æcidial form, and haustoria are numerous. They are all of the branched type: after penetrating the cell the hypha forms three or more apical branches (fig. 11). They are most numerous in the outer parenchymatous cells and chiefly in the most external ones; but they are also to be seen within the cells of the sheath and in the parenchyma cells immediately under these.

Uredo-spores.-As already stated, four patches of spore extrusion may be seen in every transverse section of a fully affected needle (fig. 23), the two larger ones being superior. In such transverse sections, it will be observed that the proportion of the circumference involved in spore formation is very large: about $\frac{8}{13}$ ths of the circumference being thus involved. The epidermal layer generally ends abruptly at some distance from the spore bed (fig. 23). There must be a considerable loss of epidermis involved in this sporing process, for the circumference of a normal needle at its middle is to so much of the circumference of an affected needle as is still covered with epidermis as 13.5 to 9.0. As regards the hypertrophy of the needles affected, it may be noted that, as the circumference of the transverse section of an affected needle at its middle was 26 units, whilst that of a normal needle at its middle was 13, the area of the former is four times that of the latter. But, as I have already stated, the specimens were not examined until they had been some days in brine, which seems to have swollen them to some slight extent.

At the bases of these spore beds, the hyphæ form as usual densely convoluted masses (fig. 25). The basidial cells composing the bases of the spore beds form a more or less irregular line of cells containing orange-red oil globules. These oil globules are more numerous in the more superficial parts. The whole thickness of the orange coloured bed is about 0.127 The basal cells which arise directly from the mycelium are large, m. m. nucleated, and of various sizes and shapes : the diameter of an averagesized cell was found to be 16μ (fig. 15 a). These cells may be stained with carmine though not brilliantly: the rest of the fungal elements do not take the stain at all. By division a row of such cells are formed (fig. 15 b.) The end cell of such a row throws out a finger like protrusion (fig. 15 c) from which the spores are separated by transverse septation. The spores (fig. 15 d) are spherical with their walls destitute of any surface markings. Their contents are granular and of an orange-red colour. The moistened spores measured on an average 9.5μ in diameter.

Such then is a very imperfect account of one of the most curious and striking of the many forms of Uredineæ which occur on the Himalayas. The fungus was first met with in May 1884, and the above account of it written immediately afterwards; but its publication was deferred in the hope that further opportunities might have been afforded me of investigating its life history. This hope I have now reluctantly abandoned, as my official engagements have not allowed me another opportunity of visiting the forests in which it occurs. The above description of its morphological characters in one stage of its existence may, however, prove of some interest and may attract the attention of others with greater opportunities and leisure than I have of following its developmental history, and is therefore published as it stands. A continued study of it is much to be desired, if only from an economic point of view, for the affection must prove very destructive to these valuable timber trees. Apart from the diversion of nutriment it must occasion, the habit it has of attacking new growing shoots and so completely involving them as to destroy them must be most injurious to these trees. I have on several occasions seen young trees (seedlings) not only with their terminal shoots involved by it, but also with many of their lateral shoots attacked at the same time.

Note.-After I had completed this paper I chanced to read, in an Appendix (B) to Hooker's "Himalayan Journals," published in 1854, of the occurrence in the Sikhim Himalayas of what may prove to be the same fungus though in some respects the description given of it does not agree with the characters of the form I have above described. It is there stated :-- "A very fine Æcidium also infests the fir tree (Abies smithiana) a figure of which has been given in the 'Gardener's Chronicle' 1852, p. 627, under the name Æcidium Thomsoni. This is allied to the Hexenbesen of the German forests but is a finer species and quite distinct." I have unfortunately not been able to refer directly to this paper, but am indebted to the kindness of a friend for the following résumé of it. The fungus was gathered by Dr. Thomson at an elevation of 8,000 feet in the Northern Himalayas on Abies smithiana. The affected leaves are reduced in length nearly one half and curved. The whole upper surface is occupied by one or more large, elevated, more or less elongated sori sometimes disposed in two rows "which must give the diseased tree a very strange appearance and at length prove fatal. The spores are greatly elongated often exceeding $\frac{1}{800}$ " in length. They were found to be mixed with mucedinous filaments some of which were ready to fructify." These were thought to be extraneous and to belong probably to some Penicillium. Dried specimens like other allied parasites had a smell of violets. The paper is very short, and the data given are not sufficient to allow of any decision being come to regarding the identity of the fungus with the form above described.

1886.]

EXPLANATION OF THE PLATES.

PLATE I.

1. Outline sketch, natural size, of three affected shoots (æcidium).

2. Needle showing fully developed æcidial fructification, about twice the natural size : æcidia in two rows on upper surface, excepting one at a on the lower surface.

3. Young æcidium emerging through a rent in the epidermis, with two spermagonia—slightly magnified.

4. Showing mode in which pseudoperidium bursts : slightly magnified.

5. Matting of mycelial filaments on surface of a parenchyma cell \times 250. Transverse section.

6. The same \times 250. Longitudinal section.

7. Æcidiospore × 580.

8. Æcidiospores with intercalary cells towards base of a series × 450.

9. Fragments of hyphæ in an air space \times 450.

10. Spermatia \times 580.

11. Haustorium in transverse section within a parenchyma cell \times 250 (uredinal affection).

PLATE II.

12. Spermagonium : transverse section \times 250.

13. a. Portions of hyphæ from a longitudinal section of needle affected with **uredinal** fungus \times 250. b. The same \times 580.

14. Convoluted mass of mycelium between parenchyma cells (uredo) \times 430. Transverse section.

15. a. Basal cubical cells with nuclei (nucleus dividing in one which springs directly from a hypha) \times 580. b. Round basal cells in series \times 580. c. Basal cells with finger-like protrusions preparatory to formation of spores \times 580. d. Free uredo-spores \times 580.

16. Peridial cells \times 340.

PLATE III (Photographs).

17. General appearance of young shoots in an early stage of infection by the secidial fungus, before the pustules have protruded through the epidermis.

18. The same when the fungus is fully developed.

19. General appearance of a shoot attacked by the uredinal fungus : fully developed.

20. Appearance of withered and dried up shoot after the death of the funges. (N. B. The four preceding figures about $\frac{2}{3}$ natural size.)

21. Transverse section of normal needle of Abies smithiana.

22. Transverse section of needle affected by æcidial fungus (the peridium with the enclosed æcidiospores has accidentally become detached).

23. Transverse section of needle affected by uredinal fungus showing four beds of spores.

24. Transverse section of needle affected by *Chrysomyxa abietis* (?) introduced for comparison with Figs. 22 and 23 as practically normal in all respects excepting the protruding fractification.

(N. B. The three preceding figures all magnified to the same extent.)

25. Transverse section of needle affected with uredinal fungus more highly magnified than Fig. 23 and illustrating several points referred to in the text; more especially the dislocation of the parenchyma cells and the comparatively undisturbed condition of the tissues within the endodermal sheath.







Parker & Coward 1th.

Mintern Bros 1mp

A. BARCLAY, Journ. As. Soc. Bengal, 1836, Vol. LV, Pt. II.

PLATE III.





23



22